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Introduction

Blood cultures are collected from patients with suspected sepsis or bacteremia. Virtually any organism may cause bacteremia. Thus, the isolation of all organisms from a blood culture must be considered significant and correlated with the clinical picture. At least 2 sets and no more than 3 sets of blood cultures should be collected from a patient with suspected bacteremia prior to the initiation of antimicrobial therapy. Collection of additional blood cultures may be indicated if the patient fails to respond to appropriate antimicrobial therapy or develops a new episode of fever or sepsis following an initial response to therapy. All sets of blood cultures received from a patient will be processed regardless of the number.

Although this section is mainly directed towards the processing of blood cultures, occasionally other specimen types (e.g. Sterile fluids, Bone marrow, abscess material) are received in blood culture bottles and thus their processing and work-up will be described in this manual.

The current blood culture system used in the Microbiology Laboratory is the Virtuo System manufactured by bioMerieux. The Virtuo tracks all bottles automatically loaded and communicating results directly to the LIS. Positive bottles are detected as growth-generated CO₂ causes a colour change in the pH sensitive disc on the bottom of the bottle.

Specimen Collection and Transport

See

Reagents/Materials/Media

See
**Procedure**

See MI_SM_RJCT to determine suitability of specimen.

A. Processing of Specimens:

See Specimen Processing Procedure MI_SM_PROC

a) Direct Examination:

Gram stain: Positive Blood Cultures Only

Acridine Orange: Positive Blood Cultures NBS, positive graph

b) Culture

<table>
<thead>
<tr>
<th>Media</th>
<th>Incubation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood Agar (BA)</td>
<td>CO₂, 35°C x 48 hours</td>
</tr>
<tr>
<td>MacConkey Agar (MAC)</td>
<td>CO₂, 35°C x 48 hours</td>
</tr>
<tr>
<td>Chocolate Agar (CHOC)</td>
<td>CO₂, 35°C x 48 hours</td>
</tr>
<tr>
<td>Fastidious Anaerobe Agar (BRUC)</td>
<td>AnO₂, 35°C x 48 hours</td>
</tr>
</tbody>
</table>

B. Interpretation of Cultures:

Negative Cultures:

i) Routine Blood, Bone marrow, sterile fluids, and general fungus/yeast cultures:

   Negative bottles are automatically resulted and discarded after 5 days incubation.
   “No growth after 5 days incubation.” Test comment: JNG@5

ii) Bone bank blood cultures:

   Negative bottles are automatically resulted and discarded after 7 days incubation.
   “No growth after 7 days incubation.” Test comment: JNG@7

iii) SBE/IE and PUO/FUO, PD Effluent, Brucella:
Negative bottles are automatically resulted and discarded after 21 days incubation.
“No growth after 21 days incubation.” Test comment: }NG21

Use the auto resulting worklist “BC Posted – No growth” to finalize all C&S cultures.
Positive Cultures:

Positive cultures are prepared in the specimen processing area.

See Specimen Processing Procedure MI_SM_PROC

Gram stain

i) Read gram stain slide from subcultured positive bottle
ii) Report smear as per Gram stain result reporting.
iii) Phone results as appropriate.

The Gram stain may indicate the need for additional media or a change in the incubation conditions. See the table below or the Charge technologist for appropriate additional media.

Additional media for preliminary processing of positive BacT/Alert blood culture bottles:

<table>
<thead>
<tr>
<th>Gram stain morphology</th>
<th>Additional Media</th>
<th>Incubation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mixed gram positive &amp; gram negative bacterial</td>
<td>2 Colistin Nalidixic Agar (CNA)</td>
<td>CO2 35°C x 48 hours</td>
</tr>
<tr>
<td></td>
<td></td>
<td>AnO₂ 35°C x 48 hours</td>
</tr>
<tr>
<td>Small gram negative bacilli</td>
<td>Campylobacter Agar (Campy)</td>
<td>Microaerophilic 42°C x 48 hours</td>
</tr>
</tbody>
</table>

Note: If a culture bottle marked as “Brucella” is flagged positive, a small amount of the blood is removed for a Gram smear ONLY. If the Gram smear shows small gram negative bacilli, forward the positive culture bottle to the Public Health Laboratory (PHOL) for identification. If the Gram smear shows organisms other than small gram negative bacilli, notify technician to proceed to subculture the bottle.

If gram stain is negative, check the bottle graph. If the graph appears to be positive, recheck and/or repeat the gram stain and/or acridine orange stain. If the graph appears to be negative, enter the gram result ”No bacteria seen” under media (GRAM). Do not assign an isolate #. Give negative bottle to technician to reload.
Sub-cultures

No growth: Read & document no growth on plates from bottles flagged positive but have no bacteria seen in gram. The culture will remain on the “BC Posted – No Iso” worklist. After the 48 hours reading of the plates and they are still no growth, press CTRL U before exiting the order to remove the “positive” flag. The order will then appear on the “BC Posted – No growth” worklist until the final BacT/Alert result is posted.

1. Identification

See Blood Culture Workflow for workflow and duties per shift.

Examine the sub-cultured plates and perform Vitek-MS or full identification as outlined in the BACTERIA AND YEAST WORKUP manual on all isolate types.

If both bottles of one set of blood cultures grow organisms with the same Gram stain result and/or have the same colonial morphology, do Vitek MS only from one bottle of the set. (*The exception to this is gram positive cocci in clusters. In this case, do Vitek MS from growth from both bottles.*)

If multiple sets from the same patient collected within 24 hours of each other are positive and growing **morphologically identical organism(s)**, perform and report complete identification from one bottle of each set (*The exception to this is gram positive cocci in clusters. In this case, do Vitek MS from growth from all bottles.*)

If yeast is isolated, do Vitek MS, using formic acid (according to Vitek MS manual). If not identified by Vitek MS, send to PHOL for Identification.

Minimum work-up is performed for identification of isolates from **autopsy blood** specimens.

1. Single isolate culture: Minimum work-up or Vitek MS

2. Mixed culture (>2): If Vitek-MS is unsuccessful, perform minimum workup, list organisms based on Gram stain morphology, growth requirement and minimum work-up e.g. “Mixed culture including Enterococci, anaerobic gram positive bacilli, aerobic gram negative bacilli……etc.”.
2. Susceptibility

Refer to Susceptibility Testing Manual.

Note: Set up susceptibilities on CNST from patients with endocarditis or if isolated from a fluid in a blood culture bottle.

If both bottles of one set of blood cultures grow the same organism, perform susceptibility on the organism from one bottle only.

If multiple sets from the same patient collected within 24 hours of each other are positive and growing identical organism(s), susceptibility testing can be referred.

If multiple sets from the same patient collected within 7 days of each other are positive and growing identical yeast organisms, susceptibility testing can be referred.

For isolates of *Staphylococcus aureus* or *Enterococcus* one bottle from each set of BC bottles must have an oxacillin and/or vancomycin screen performed.

No sensitivity is required for autopsy blood or bone bank specimens.

**Blood Culture Isolates to be Frozen and Saved**

- Freeze ALL isolates from blood culture including Autopsy isolates at -70°C EXCEPT:
  - Enterococcus susceptible to Vancomycin
  - Anaerobes
  - *E. coli* susceptible to Amp and Septra
  - Skin flora (CNST, Micrococcus, *Bacillus* sp., *Corynebacterium* sp. not JK, *Lactobacillus* sp., *Lactococcus* sp., *Propionibacterium* spp., *Peptostreptococcus* sp.)
  - Repeat isolates within 24 hours
  - Document freezing in Softstore.
Reporting Results

Negative to Date Bottles and False Positive Bottles:

Preliminary: The LIS will automatically report “Culture received in lab. Results will be reported as soon as they become available” and assign a preliminary status.

Negative report:

Final: i) Routine Blood, Bone marrow Sterile Fluids, Blood products, Fungus, Yeast "No growth after 5 days incubation”.

ii) Bone Bank bloods “No growth after 7 days incubation”.

iii) Brucella, PD Effluent, SBE / IE, PUO / FUO “No growth after 21 days incubation”.

iv) Dimorphic fungi “No fungus isolated”. (See Mycology Manual)

Positive report:

For Gram stain results:

In LIS “MEDIA” Window, under GRAM media, pick from keypad:
1. The bottle type the organism was from e.g. from FO2.
2. Then pick the organism seen e.g. gram positive cocci in clusters
3. Then the isolate code to be transferred to the “ISOLATE” Window (if this is the first time this organism is seen in this order; omit this keypad pick if this is the second time this organism is seen in this order.)

Go to the “TEST” Window:
1. For Blood Culture test, REMOVE preliminary statement “Culture received in lab…” Add “UPDATED REPORT”.
2. For fluids or aspirates in blood culture bottles report Gram results under the “ISOLATE” window of LIS as Isolates 1 "Gram positive cocci” etc. REMOVE
preliminary statement “No growth to date……..” and add “UPDATED REPORT” and Status the test (C&S or FLDM) as preliminary (^P).
No work has to be documented under test code ‘?BTLE’

3. Go to the “ISOLATE” Window.
4. Go to the ISOLATE COMMENT field.
5. From isolate keypad, select “BLDC” to go to Blood Culture specific keypad.
6. Select >SMEAR, then select the appropriate comment for the organism morphology and “seen” e.g. “~in clusters seen”.
7. Press “Verify all”
8. Save the isolate and return to “TEST” window.
9. Status the Test as preliminary (^P).

For all sites, telephone the ward/ordering physician as soon as the Gram stain result is available. If another bottle of the same set becomes positive with the same organism, no further report is required. Infectious Disease Team (ID) must be paged for new yeast seen.

**For Culture results:**
Remove “~seen” comment from ISOLATE COMMENT field and add “isolated”

Report organism with the corresponding antibiotic susceptibilities results and comments as appropriate, refer to Susceptibility Manual and Vitek MS Species List.

For identification and susceptibility results, call the results to the wards as soon as they become available as follows:

<table>
<thead>
<tr>
<th>Hospital</th>
<th>Monday - Friday</th>
<th>Weekend / Holidays</th>
</tr>
</thead>
<tbody>
<tr>
<td>TGH</td>
<td>No call*</td>
<td>No call*</td>
</tr>
<tr>
<td>TWH</td>
<td>No call*</td>
<td>No call*</td>
</tr>
<tr>
<td>TRI</td>
<td>No call*</td>
<td>No call*</td>
</tr>
<tr>
<td>PMH</td>
<td>No call*</td>
<td>Call</td>
</tr>
<tr>
<td>MSH all wards and patients admitted from Emergency ward</td>
<td>No call*</td>
<td>No call*</td>
</tr>
<tr>
<td>MSH Emergency Ward not admitted or discharged</td>
<td>Call</td>
<td>Call</td>
</tr>
<tr>
<td>Bridgepoint,</td>
<td>No call*</td>
<td>No call*</td>
</tr>
<tr>
<td>Baycrest</td>
<td>No call*</td>
<td>No call*</td>
</tr>
<tr>
<td>CAMH</td>
<td>Call</td>
<td>Call</td>
</tr>
<tr>
<td>Grace</td>
<td>Call</td>
<td>Call</td>
</tr>
</tbody>
</table>

*Unless a new organism is isolated that was not seen on the initial Gram stain, or the organism has been identified as *Streptococcus pneumoniae*, *Listeria monocytogenes*,

NOTE: This document is Uncontrolled When Printed.
Any documents appearing in paper form that are not stamped in red "MASTER COPY" are not controlled and should be checked against the document (titled as above) on the server prior to use.
Management System\UHN_Mount Sinai Hospital Microbiology\Standard Operating Procedures\Bacteriology Procedures\
Staphylococcus aureus, Streptococcus pyogenes, Neisseria meningitides, Salmonella species or Cryptococcus neoformans.

When both bottles in the set are completed, assign “Interim” status (^L). Senior staff will review and finalize the report.

**Infection Control Team Reporting:**
Notify as per QPCM116003

**Infectious Diseases Team Reporting:**

If *S. aureus*, Yeast or Fungus is isolated (presumptive or confirmed) from an inpatient at the TGH, TWH, PMH or MSH, the relevant Infectious Disease (ID) Team must be notified immediately if the hours are 0800-midnight. Notification of positives to ID team from midnight-0800 should be deferred until the next morning.

For patients whose blood was drawn in the emergency department, call ID only if the patient is admitted.

**DO NOT** notify the Infectious Disease team if the patient is deceased, was seen in an outpatient clinic, discharged from the emergency department.

Notify the relevant Infectious Disease team by paging the on-call physician covering the team through locating as follows:

**A. Positive Blood Cultures for *S. aureus*, yeast, or fungus 0800-midnight:**

<table>
<thead>
<tr>
<th>Hospital</th>
<th>Infectious Disease (ID) Team (Monday-Friday):</th>
<th>Infectious Disease (ID) Team (Weekends/Holidays):</th>
<th>Locating Phone Number</th>
</tr>
</thead>
<tbody>
<tr>
<td>TWH</td>
<td>TWH Infectious Disease</td>
<td>Tri-hospital Infectious Disease</td>
<td>14-3155</td>
</tr>
<tr>
<td>PMH</td>
<td>PMH Immunocompromised ID</td>
<td>Tri-hospital Infectious Disease</td>
<td>14-3155</td>
</tr>
<tr>
<td>MSH NICU</td>
<td>Sick Kids Infectious Disease*</td>
<td>Sick Kids Infectious Disease*</td>
<td>416-813-7500</td>
</tr>
<tr>
<td>MSH not NICU</td>
<td>Tri-hospital Infectious Disease</td>
<td>Tri-hospital Infectious Disease</td>
<td>14-3155</td>
</tr>
<tr>
<td>TGH Transplant floors (7MA or TGH)</td>
<td>TGH Immunocompromised ID</td>
<td>Tri-hospital Infectious Disease</td>
<td>14-3155</td>
</tr>
</tbody>
</table>
B. Positive Blood Cultures for *S. aureus*, yeast, or fungus midnight-0800:

No calls to any ID service during these hours. Call the next morning following the table above.

**BC Bench Pending Worklists**

Pending worklists have been created to ensure all orders are accounted for. Technologist work-up benches must check the following pending lists daily:

**Pending list 1: BC posted - No Isolate Worklist**

This worklist includes all orders which had a positive bottle but do not have an isolate. Check all orders in this worklist:
- Check if bottle is positive but NBS. These are okay and will remain on this list.
- Check to ensure all positives bottles with an organism seen in the gram stain have an isolate entered. This is to ensure a gram smear is not lost or missed for a new positive bottle.

**Pending list 2: BC posted - with ISO**

This worklist includes all positive orders with isolates being worked on. It will catch orders that were NOT assigned to a bench worklist. For orders on the list not assigned to a bench:
- Check the list for completeness of workup (culture not missed).
- Check orders for second bottles now negative that can be interim’d.
Pending list 3: BC bench

Each blood culture bench has a worklist of all orders being processed on that particular bench.

- Review the list for completeness of workup (culture not missed).
- Check orders for second bottles now negative that can be interim’d.
  - 5 days = 120hrs
  - 7 days = 168hrs
  - 14 days = 336hrs
  - 21 days = 504hrs

Reference


CMR 1995 8(4):447-483 and QMPLS Broadsheet on ESBL and ampC Resistance in GNB (updated 2007-12-10)
ISOLATER 10 BLOOD CULTURE SYSTEM FOR DIMORPHIC FUNGI

Introduction

The Isolator 10 blood culture system should be used for the isolation and detection of Cryptococcus and dimorphic fungi such as Histoplasma and Blastomyces.

If BacT/Alert bottles are received with a request for dimorphic fungi, notify the ward / ordering physician that they must use the Isolator 10 collection tubes. The BacT/Alert bottles should only be processed as per routine blood cultures.

Collection and Transport

See Specimen Processing Procedure MI_SM_PROC

Reagents / Materials / Media

See Analytical Process - Bacteriology Reagents/Materials/Media List QPCMI10001

Procedure

A. Processing of Isolator 10 Microbial Tubes:

See Specimen Processing Procedure MI_SM_PROC

B. Interpretation of Fungal Culture Plates:

Refer to.

Reporting Results

Refer to Mycology Manual..

Reference

1. Isolator 10 Product Insert.
APPENDIX I - Quality Control (QC) of the Virtuo System

Daily

When entering the “Blood culture Posted-No Iso” worklist, the LIS will prompt “QC pending: Would you like to bridge to QC?” Enter “Y”.

i) Check the Bone Bank report to be sure all Bone Bank bloods have been edited to a maximum test time of 7 days.

ii) Check the PD effluent report to be sure all have been edited to a maximum test time of 21 days.

iii) Check the O/E Comment Report to look for orders from the previous day that may have comments that were missed when loaded. Look for requests for:

- Brucella - bottle should be labeled “Brucella”.
- Specific requests for Dimorphic fungi (e.g. Histoplasma, Blastomyces) – should be collected in an Isolator tube
- SBE/IE, FUO/PUO – keep 21 days
APPENDIX II - Sterility Testing of Blood for Media

Initial testing (performed by blood culture bench)

On receipt in the laboratory, each bottle is assigned a letter (A, B, C, etc.). Aerobic BacT/Alert bottles are labelled with corresponding letters. The smaller portion of the barcode is attached to the original bottle of blood.

Enter the data in the LIS as follows:

- **MRN**: 77777777777
- **TESTS**: BLOOD CULTURES
- **SOURCE**: BFA
- **SITE**: HORSE / SHEEP, LOT #, EXP DATE

With a needle and syringe, 2.5 mL of blood is aseptically transferred from each bottle of blood and inoculated into separate BacT/Alert bottles. The original bottles of blood are immediately refrigerated and the BacT/Alert bottles loaded and processed as routine specimens.

If any BacT/Alert bottle gives a positive reading, the QA technologist must be informed ASAP and the original bottle of blood is removed from use. The BacT/Alert bottle is Gram stained and subcultured to BRUC (Ana2) and CHOC (CO2). Identification to the species level (e.g. staphylococcus, diphtheroid, etc.) will be sufficient.

After Use (performed by media preparation and the QA technologist)

As each bottle of blood is used, the last few drops of blood are inoculated onto a BA plate which is labelled with the lot # and letter. This plate is incubated at 35°C for 48 hours, then at RT for 48 hours. The results are recorded as a QC item in the LIS for the medium that the blood has been added to.
APPENDIX III - Daily Virtuo System Checks

Daily checks

Order Entry Comments daily printouts:
- Check comments for query Brucella, SBE/IE,PUO/FUO.
- Ensure cultures have “MAX TEST TIME” edited to 21 days under “Bottle Data” in DATA MANAGER.
To change incubation times refer to Changing Incubation Times

Anonymous and Orphan bottles - bottles missing patient data:
- At instrument home screen, select “Reports” icon and select “Orphan” / “Anonymous” icon. If no items appear on the list there are no problems.
- For items appearing on the “Orphan” list see Troubleshooting section for Orphan Bottles.
- For items appearing on the “Anonymous list” see Troubleshooting section for Anonymous Bottles.

Virtuo LIS Worklists

Blood Culture Receiving Working
- Check all UHN/Baycrest/Bridgepoint blood cultures not received in the laboratory after 48 hours. Phone ward and fully investigate missing bottles.
- Check all other blood culture orders not received in the laboratory after 7 days. Final orders using the R (Reject) keypad and selecting the comment }NR7D.

BC Not Received
This worklist contains blood cultures that were collected but not yet received in the microbiology.
- Check all orders not received >48hrs after collection.
- Check Myla for received time and date and document in LIS order
- Follow up any orders not in Myla with the ward.

BC Cancelled (by HIS)
- Search all orders cancelled by HIS in Myla for received time and date.
- In order entry “add next” to create a new order with original collection and received information.
• Document in result entry on the new order, the LIS# of the cancelled order.
• Locate bottles in Virtuo, unload and affix new order labels. Before reloading, edit each bottle of the set with the new accession number and save.
• Reload bottles in the same controller that they originated.

**New fluid in Bottle**
• Prelim all orders with “~No growth to date…” comment after overnight incubation
  o from main test keypad select “p” for prelim comments.

**PD Effluent NEW (21 days)**
• Prelim all orders with “~No growth to date…” comment after overnight incubation
  o from main test keypad select “p” for prelim comments.
• Change incubation of bottles in Virtuo to 21 days
  o Document date you changed the incubation under media BC21

**BC ENACT NEW (14 days)**
• Prelim all orders with “~No growth to date…” comment after overnight incubation
  o from main test keypad select “p” for prelim comments.
• Change incubation of bottles in Virtuo to 14 days
  o Document date you changed the incubation under media BC14
  o On the BCBC line in “R” column to add a red Checkmark (double click)
APPENDIX IV - Troubleshooting the Virtuo System

Troubleshooting

The Virtuo system is equipped with alert system that notifies attention is required by an audible alarm and green flashing light on the top at the top of the related Virtuo instrument.

Virtuo Troubleshooting Guide is available on the Virtuo main screen.

For service call bioMerieux at 1-800-361-7321

The following is a guide for troubleshooting common Virtuo alerts with the use of worklists and checks.

Changing Incubation Times

Follow this procedure for any specimens which greater than 5 days incubation has been requested:

In the LIS result entry, on the line for BCBC
- Right click the “R” on the right hand of the line and select “Result Media” or, double click in the spot to make the red check mark appear.
- From BCBC media, add new media BC21 media
- Under BC21 record the date (date incubation was modified in Virtuo)
- Record the auxiliary number (UHN) or LIS number (for all other hospitals)
- Take this number to the Virtuo Instrument and locate the specimen using the auxiliary number (UHN) or LIS number (for all other hospitals)
- Under SEARCH, key in the order number, VIEW, EDIT, and change the incubation time

*You will need to check in LIS for all specimens currently incubating in Virtuo for that patient and do this process for each one

Orphan Bottles

These are bottles missing patient data often due to unreadable patient barcodes.
- Select “Resolve” from the main screen of the alarming unit.
- An image of the label will appear on screen.
  - DO NOT attempt to input the missing data using the keypad.
• Choose “Unload Bottle” icon
• Inspect bottle to see why barcode did not read.
• If Scannable:
  o Scan the label manually into the virtuo missing field
• If not Scannable
  o Reprint a label using the LIS, confirming correct patient.
  o Scan the label into the virtuo missing field
  o Click “Save” and reload bottle in controller it originated from.

For bottles with auxiliary numbers attached:
• Check EPR for orders not accepted within EPR
• For orders not found in EPR see LIS or Senior tech.

**Anonymous Bottles**

These are bottles missing bottle data often due to unreadable patient barcodes.

• Select “Resolve” from the main screen of the alarming unit.
• An image of the label will appear on screen.
  o Type the bottle number from the photograph into the bottle ID field

Alternatively if bottle label cannot be read:

• For bottles loaded within 10 minutes:
  o Unload and remove patient label obscuring bottle ID.
  o Scan bottle barcode and save.
  o Reload bottle in same controller it originated from.
  o If unable to have barcode visible, place a generic label on the bottle and scan into the virtuo. **Ensure to select bottle type.**
  o Reload bottle in same controller it originated from.
• For bottles loaded for greater than 10 minutes or an unknown amount of time:
  o DO NOT UNLOAD. All bottle readings will be lost.
  o Scan a generic label into the Virtuo. **Ensure to select bottle type.**
  o Save.
  o It is now safe to unload the bottle.
  o Affix the generic label onto the bottle and reload into same controller it originated from.
### Record of Edited Revisions

**Manual Section Name: Blood Culture Manual**

<table>
<thead>
<tr>
<th>Page Number / Item</th>
<th>Date of Revision</th>
<th>Signature of Approval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Annual Review</td>
<td>May 30, 2001</td>
<td>Dr. T. Mazzulli</td>
</tr>
<tr>
<td>Annual Review</td>
<td>May 30, 2002</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>Annual Review</td>
<td>May 30, 2003</td>
<td>Dr. T. Mazzulli</td>
</tr>
<tr>
<td>Page 15, reporting of <em>S. aureus</em> to ID resident</td>
<td>July 27, 2004</td>
<td>Dr. T. Mazzulli</td>
</tr>
<tr>
<td>Handling of special request for <em>Brucella</em></td>
<td>December 17, 2004</td>
<td>Dr. T. Mazzulli</td>
</tr>
<tr>
<td>Reload all false positive bottles regardless of the number</td>
<td>December 17, 2004</td>
<td>Dr. T. Mazzulli</td>
</tr>
<tr>
<td>of times flagged.</td>
<td></td>
<td></td>
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<tr>
<td>See Bacteria work-up manual for isolate work-up</td>
<td>December 17, 2004</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>Refer to previous isolate up to 72 hours; same with <em>Enterococcus</em></td>
<td>December 17, 2004</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>Refer to previous isolate up to 72 hours for freezing.</td>
<td>December 17, 2004</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>Do not report the number of bottles positive</td>
<td>December 17, 2004</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>New troubleshooting Reports 6 and 7</td>
<td>December 17, 2004</td>
<td>Dr. T. Mazzulli</td>
</tr>
<tr>
<td>Remove subculture on SAB for yeast page 7</td>
<td>December 17, 2004</td>
<td>Dr. T. Mazzulli</td>
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<td>December 17, 2004</td>
<td>Dr. T. Mazzulli</td>
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<td>CHC/Ajax No need to call ward with sensitivities and ID Page 12</td>
<td>April 13, 2005</td>
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<td>Call ID physician including <em>S. aureus</em> and SPICE bugs</td>
<td>September 21, 2005</td>
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<td>Blood collection procedure</td>
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<td>UHN Bone Marrow accessioning</td>
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<td>Set up DENKA as soon as presumptive ID of <em>S. aureus</em> and</td>
<td>February 14, 2007</td>
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<td>enough growth</td>
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<td>CTRL U for false positives to remove LIS Positive flag</td>
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<td>Dr. T. Mazzulli</td>
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<td>Modify Mycology plates for ISOLATOR 10</td>
<td>June 29, 2007</td>
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<td>Do not need to call ID and sensi to MSH wards</td>
<td>May 16, 2008</td>
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<td>Changed reporting positive gram as “isolate”</td>
<td>January 30, 2008</td>
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<td>Added SPICE bug reference and remove Cedecea from SPICE list</td>
<td>January 30, 2008</td>
<td>Dr. T. Mazzulli</td>
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<td>CNST Sensi change</td>
<td>February 13, 2008</td>
<td>Dr. T. Mazzulli</td>
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<td>Appendix VI - Ajax Blood Culture processing changed to RVHS Blood culture Processing</td>
<td>April 01, 2008</td>
<td>Dr. T. Mazzulli</td>
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<td>Discharged ER patients with S. aureus or SPICE bugs – stop call to ID.</td>
<td>May 16, 2008</td>
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<td>Change PD fluid incubation to 21 days per Hemodialysis Unit request</td>
<td>June 1, 2009</td>
<td>Dr. T. Mazzulli</td>
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<td>Positive blood with yeast; add direct germ tube</td>
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<td>Dr. T. Mazzulli</td>
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<tr>
<td>Positive blood with yeast or fungus, page ID resident/physician</td>
<td>July 24, 2009</td>
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<td>Annual Review</td>
<td>June 1, 2010</td>
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<td>Expanded TDNA reading instructions</td>
<td>November 23, 2011</td>
<td>Dr. T. Mazzulli</td>
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<td>Annual Review</td>
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<td>Dr. T. Mazzulli</td>
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<td>Clarify Denka refer back, page 9</td>
<td>October 9, 2012</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>Removed Appendix VI – Processing RVHS blood Cultures</td>
<td>October 9, 2012</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>Updated Autopsy Blood work-up, reporting statement</td>
<td>December 05, 2012</td>
<td>Dr. T. Mazzulli</td>
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<td>Annual Review</td>
<td>December 05, 2012</td>
<td>Dr. T. Mazzulli</td>
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<td>CNST reporting phrase changed</td>
<td>May 13, 2013</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>CNST reporting phrase changed back</td>
<td>July 16, 2013</td>
<td>Dr. T. Mazzulli</td>
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<td>Removed CNST not S. lugdunensis susceptibility testing Set up susceptibilities on CNST in BC if isolated from patients with endocarditis</td>
<td>November 10, 2013</td>
<td>Dr. T. Mazzulli</td>
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<td>CNST reporting phrase changed</td>
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<td>Annual Review</td>
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<td>Change to work up to MS</td>
<td>March 28, 2014</td>
<td>Dr. T. Mazzulli</td>
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<td>Freeze ALL isolates from blood culture including Autopsy isolates</td>
<td>March 28, 2014</td>
<td>Dr. T. Mazzulli</td>
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<td>Annual Review</td>
<td>March 28, 2014</td>
<td>Dr. T. Mazzulli</td>
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<td>BC positive for S. aureus and yeast/fungi should be called to Sick Kids ID as of Aug 1, 2014</td>
<td>July 20, 2014</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>Host Not Responding Message</td>
<td>August 9, 2014</td>
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### Table of Changes

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<tr>
<td>- Autopsy blood: -Single isolate: minimum work up or MS &amp; specified Mixed culture is (\geq) or equal to 2. -False positive cultures: - Moved last paragraph to work up of false positive (\rightarrow) culture (\sim) p.23 (this section is work up of initial pos bottle accessioning bloods side) -Processing of sub-culture: -successful ID from maldi is GREATER than or equal to 98% -Removed susceptibility comments; refer to manual and vitek ms isolate manual -removed reminder for DENKA, refer to sensitivity manual. - Updated AT LIS section of download problem with patient demographics. Removed section: APPENDIX V - Handling of Bone Marrow in Blood Culture Bottles from UHN is gone</td>
<td>September 10, 2014</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>Revised sections for:</td>
<td>December 1, 2014</td>
<td>Dr. T. Mazzulli</td>
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<td><em>For Gram stain results</em></td>
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<td><em>For Culture results</em></td>
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<td><em>Infection Control Team Reporting</em></td>
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<td><em>Infectious Diseases Team Reporting</em></td>
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<td><strong>Revised: Infectious Diseases Team Reporting:</strong> (p.14)</td>
<td>March 4, 2015</td>
<td>Dr. T. Mazzulli</td>
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<td>Reorganize</td>
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<td>March 11, 2015</td>
<td>Dr. T. Mazzulli</td>
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<td>p.16 Modified Isolater processing to : Centrifuge blood at 3000g</td>
<td>November 12, 2015</td>
<td>Dr. T. Mazzulli</td>
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<td>Positive cultures: additional media added incubation criteria to table</td>
<td>March 4, 2016</td>
<td>Dr. T. Mazzulli</td>
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<tr>
<td>Manual bottle procedure modified to include further macroscopic examination:</td>
<td>June 16, 2016</td>
<td>Dr. T. Mazzulli</td>
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<td>Examine all bottles for macroscopic growth twice daily for day 1 and day 2, record results in the LIS. For the remainder of the incubation period check bottles macroscopically at least daily and record in the LIS</td>
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<tr>
<td>Annual Review</td>
<td>February 23, 2017</td>
<td>Dr. T. Mazzulli</td>
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<td>Processing of positive BC moved to Specimen</td>
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<th>Processing Manual. Updated procedures from BacTAlert to Virtuo procedures.</th>
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</table>
| Updated Error reports for the Virtuo Added Report:  
- Bottles with missing patient information | March 7, 2017 | Dr. T. Mazzulli |
| Removed Reports:  
- Bottle with wrong accession #  
- Negative to Date unloaded by mistake  
- Loading problem - bottle ID without accession number  
- Accession number with wrong bottle | | |
| Added Virtuo Troubleshooting appendix with customer service information, and appendix for worklists/daily checks. Removed appendix I QC of Virtuo system, information added to BC manual under section BC Bench Pending Worklists and troubleshooting appendix. | May 8, 2017 | Dr. T. Mazzulli |